

# FINAL TECHNICAL REPORT

## OPTICAL CROSS-SECTIONS OF MARINE PARTICLES

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### *REPORT FOR THE FISCAL YEAR 1993*

#### LONG-TERM GOALS

The long-term goal of this research is to understand the magnitudes and variability of the optical properties in the upper ocean and to derive mathematical descriptions that will allow predictions of spectral absorption and scattering by sea water.

#### OBJECTIVES

Our near-term objective is to conduct a comprehensive study of the optical properties of marine microorganisms and other biogenic particles in order to determine their optical cross-sections and contribution to absorption and scattering by sea water. Our major effort during the last year was aimed at quantifying diel changes in scattering and absorption cross-sections of phytoplankton cells. This study is related to our hypothesis that the relatively large diel cycle in the beam attenuation coefficient that has been observed in the ocean is in part caused by diel change in the size and refractive index of cells comprising the phytoplankton community.

Another effort was focused on testing the applicability of the dynamic light scattering (DLS) technique for characterizing the size distribution of small marine particles ( $< 5 \mu\text{m}$ ) including the submicron range. While we previously used this technique for studying the cultures of heterotrophic marine bacteria, this time we focus our attention on natural assemblages of marine particles. This effort is related to the hypothesis that submicron particles make major contribution to particulate backscattering in the ocean.

#### APPROACH

We examine the microorganisms or microparticles of interest by growing them in the laboratory or isolating and concentrating them in the field. We then measure both the absorption and beam

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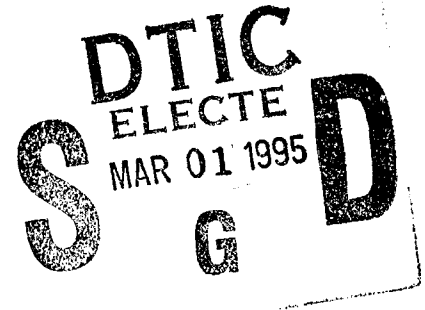
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attenuation coefficients of the suspensions with, and measure the size of the particles with either a microscope or an electronic particle counter. From these measurements, we determine the attenuation, absorption, and scattering cross-sections and efficiency factors which represent a "mean" particle derived from a given assemblage. We then determine the refractive index of particles by solving the inverse problem based upon Mie scattering theory. This allows us to calculate the volume scattering function and backscattering properties of particles. In order to study diel changes in the optical properties of phytoplankton cultures, the laboratory experiments are designed specifically to include frequent sampling for optical and cell size measurements, so that the variations in scattering and absorption by cells on hourly time scales can be resolved. In order to characterize the size of small marine particles we apply the DLS technique. The underlying principle of this technique is that the net intensity of light scattered from a collection of small particles fluctuates with time. This is the result of interference of the light scattered by particles, whose position in the region illuminated by laser changes continuously due to various motions including Brownian motion.

## TASKS COMPLETED

Laboratory experiments were conducted to study diel variations in the optical properties of the marine diatom *Thalassiosira pseudonana*. This centric diatom was chosen to represent eukaryotic phytoplankton cells. The examined cells have a size (ca. 4  $\mu\text{m}$ ) similar to many oceanic nanophytoplankters that are known to contribute significantly to the bulk optical properties of the upper ocean. We also completed the analysis of data from our previous experiments which were designed to examine the effect of light fluctuations induced by surface-wave focusing on the optical properties of marine phytoplankton. In addition, we conducted experiments with samples of natural seawater, in which we applied the DLS technique. We described an inverse method for estimating the general slope of size distribution of small particles from the average hydrodynamic diameter as determined from the DLS measurements.

## RESULTS

Our study of the marine diatom cultures shows that the optical cross-sections of phytoplankton cells can vary greatly over a diel cycle (Stramski and Reynolds, 1993). The absorption cross-section increased by as much as 35%, and the attenuation and scattering cross-sections by 80% between minima near sunrise and maxima near sunset. These changes were associated with significant variations in refractive index and size of cells. Diel changes in the imaginary part of the refractive index accounted for 58-73% of the difference between the daily maxima and minima in the absorption cross-section. Variations in the real part of the refractive index contributed substantially (up to 85%) to the daily range in the scattering and attenuation cross-sections. Increases in cell size were important in the morning, when their contribution to changes in the optical cross-sections was similar to that associated with refractive index. In addition, the normalization of attenuation or scattering cross-sections to cellular carbon content resulted in a threefold reduction in the magnitude of diel variations.

The study of the marine chlorophyte *Dunaliella tertiolecta* grown under fluctuating light caused by surface-wave focusing shows that the size, refractive index and optical properties of cells are

not affected by whether the incident irradiance is fluctuating or constant (Stramski et al., 1993). This conclusion is valid for *Dunaliella* cells adapted to high levels of irradiance.

The results from our dynamic light scattering measurements of seawater samples are encouraging, and suggest that the general slope of the size distribution (assumed to be a power function) of small particles can be derived from the average hydrodynamic diameter of particles (Stramski and Sedlak, 1994). In addition, the multi-angle DLS measurements can be used to obtain useful information about representative value of the refractive index of particles.

## ACCOMPLISHMENTS

Our major recent accomplishment is that for the first time we provided direct evidence that the bulk optical properties of a phytoplankton suspension may undergo significant diel variations independent of the numerical concentration of cells. This has important implications for optical studies in the ocean, and will help us better interpret optical variability in the ocean. Specifically, we have to take into consideration the fact that varying particle concentration need not be the only (or even major) factor responsible for diel variations of the optical properties in the upper ocean; changes in the refractive index and size of phytoplankton cells may actually be very important, especially in situations when cell division is synchronized to some degree by the light-dark cycle. We also successfully applied the dynamic light scattering method to the analysis of natural seawater samples. This accomplishment is important because it represents a new approach to the study of submicron marine particles which are difficult to examine by commonly used techniques such as microscopy and electronic counters.

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## ***REPORT FOR THE FISCAL YEAR 1994***

### **LONG-TERM GOALS**

The long-term goal of this research is to develop the base of knowledge necessary to understand the magnitudes and variability of the optical properties in the upper ocean, and to derive mathematical descriptions that will allow predictions of spectral absorption and scattering by sea water, given the types and concentration of suspended particles.

### **OBJECTIVES**

Our near-term objective is to conduct a comprehensive study of the optical properties of marine microorganisms and other biogenic particles in order to determine their optical cross-sections and contribution to absorption and scattering by sea water. Our major effort during the last year was aimed at examining diel changes in scattering and absorption cross-sections of marine cyanobacteria which represent small autotrophic organisms from the picoplankton size range ( $\sim 1 \mu\text{m}$  in size). This work was designed to supplement our previous study of larger phytoplankton cells. The research on diel variability is related to our hypothesis that the relatively large diel cycle in the beam attenuation coefficient that has been observed in the ocean is in part caused by variations in the refractive index and size of cells comprising the phytoplankton community.

Another objective was to assess the effect of gas microbubbles on light scattering in the ocean. This effort stems from our pursuit of a comprehensive description of a budget for light scattering in the upper ocean. We believe that such a description should include not only various types of particles and microorganisms but also gas microbubbles. Such scattering objects are always present in the near-surface ocean as evidenced by observations in quiescent seas. The presence of persistent populations of microbubbles in the upper ocean may be largely associated with biological processes.

### **APPROACH**

Our approach that involves both experimental and theoretical efforts is to examine the microorganisms of interest by growing them in the laboratory. We measure the absorption and beam attenuation coefficients of cell suspensions using a spectrophotometer with a special geometrical configuration. These measurements are usually done in the spectral region from 350 to 750 nm with 1-nm resolution. We also measure the concentration and size distribution of particles in suspension with a microscope and electronic particle counter. From these measurements we determine the attenuation, absorption, and scattering cross-sections and efficiency factors which represent a "mean" particle derived from a given assemblage. We then estimate the complex refractive index of particles by solving the inverse problem based largely upon Mie scattering theory. This finally allows us to calculate the volume scattering function and backscattering properties of particles from this theory.

During the last year, in order to study diel changes in the optical properties of phytoplankton, the laboratory experiments were designed specifically to include frequent sampling for optical and cell size measurements, so that the variations in scattering and absorption by cells on hourly time scales could be resolved. To accomplish the other objective related to the influences of microbubbles on ocean optics, Mie calculations were employed to determine scattering efficiency of different-sized air bubbles suspended in water. These results were combined with literature data on the concentration and size distribution of bubbles in quiescent seas to assess the contribution of bubble populations to the scattering and backscattering coefficients of sea water.

## TASKS COMPLETED OR TECHNICAL ACCOMPLISHMENTS

Laboratory experiments were conducted to study variations in the optical properties of the oceanic unicellular cyanobacterium *Synechococcus* grown under a day-night cycle in natural irradiance. These tiny microorganisms were chosen to represent a prominent component of oceanic prokaryotic autotrophs which can significantly influence the optical properties of sea water. This is the second time when such a specific experiment on diel optical variability has been accomplished; the first one was also conducted in our laboratory but with larger eukaryotic phytoplankters. The analysis of data from the diel experiment was completed which reveals the variations in the optical cross-sections of *Synechococcus* cells and the significance of changes in the cell refractive index and size over a day-night cycle. In addition, Mie scattering calculations of light scattering by an air bubble in water and the analysis of the contribution of persistent populations of microbubbles on light scattering by sea water were performed.

## RESULTS

Our study of the cyanobacteria cultures shows that the optical cross-sections of such cells vary greatly over a day-night cycle, showing a minimum near dawn or midmorning and maximum near dusk (Stramski et al., 1994). During daylight hours, the scattering ( $\sigma_b$ ) and attenuation cross-sections ( $\sigma_c$ ) may increase even more than two-fold, and the absorption cross-section ( $\sigma_a$ ) by as much as 45%. The real part of the refractive index ( $n$ ) and size of the cells exhibited a tendency to increase during the light period and decrease at night. Changes in  $n$  had equal or greater effect than the varying size distribution on changes in  $\sigma_b$  and  $\sigma_c$ . The imaginary part of the refractive index showed generally little variation over a day-night cycle, and  $\sigma_a$  was largely driven by variations in cell size. The real part of the refractive index was correlated with the intracellular carbon concentration, and the imaginary part with the intracellular chlorophyll *a* concentration. The carbon-specific attenuation of *Synechococcus* also showed significant diel variability with an increase during daylight hours. Because we previously observed similar diel variations in the optical cross-sections of eukaryotic cells representing centric diatoms, we now suggest that such diel patterns may be a general characteristic of marine phytoplankton.

The assessment of the optical role played by microbubbles was made assuming various concentrations and size distributions of bubbles in the size range from 20 to 300  $\mu\text{m}$  (Stramski, 1994). These assumptions were made to represent typical persistent bubble populations in surface waters during nearly calm sea conditions. The results suggest that the contribution of these bubbles to the scattering and backscattering coefficients cannot be neglected in some oceanic

situations. This contribution varies between a fraction of 1% and several percent, and might possibly be as high as a dozen or so percent. Because no in situ data about microbubbles less than 20  $\mu\text{m}$  in size are available, these small bubbles are not taken into account in this assessment. Therefore, the present estimates of the influence of microbubbles on light scattering in quiescent seas are conservative.

## IMPACT FOR SCIENCE OR SYSTEMS APPLICATIONS

Our research on diel variability in phytoplankton cells has important implications for the science of ocean optics. First, our accomplishments in this area will help us better understand and interpret optical variability in the ocean, which includes the information acquired from shipboard instruments, moorings and satellites. Specifically, the impact of our findings is to change the generally accepted notion that optical variability such as diel changes in the beam attenuation coefficient are well explained by variations in particle concentration alone. Our study provides strong evidence that varying particle concentration need not be the only (or even major) factor invoked to explain diel variations of the optical properties in the upper ocean; changes in refractive index and size of phytoplankton cells over a day-night cycle may actually be very important. In addition, the study of diel variations of the optical properties of phytoplankton is important because it was recently proposed that oceanic primary productivity can be estimated from measurements of diel variability in the beam attenuation coefficient. The impact of our results in this context is two-fold. First, we caution against uncritical estimation of particle production rates from such a diel signal because changes in the optical cross-sections of phytoplankton cells may significantly contribute to the beam attenuation signal in many oceanic situations. Second, we similarly believe that the use of the diel signal in the beam attenuation for estimating primary productivity in terms of organic carbon cannot just be based on the assumption that the carbon-specific attenuation coefficient of phytoplankton is constant.

Our study of light scattering by gas microbubbles is important because a better knowledge of the optical role played by each and every one optically significant component of sea water is prerequisite to a good understanding of marine optical environment. The question of what effect the persistent bubble populations might have on ocean optics has not received, to our knowledge, any significant attention in the past. Our study is thus a first attempt to address this question. We believe it will provide inspiration for further exploration as we suggest that such microbubbles have non-negligible contribution to light scattering in this ocean.

## TRANSITIONS ACCOMPLISHED AND EXPECTED

None to date on this project but we anticipate that this work will lead to creation of a catalog of the optical properties of marine particles and will thus provide much needed information for predictive optical models used by the US Navy.

## RELATIONSHIP TO OTHER PROJECTS

The information on the optical properties of microbial particles acquired in this work is used in another ONR-sponsored project which is aimed at studying influences of microbial particles on

oceanic optics (Mobley and Stramski, 1994). In that project, we take advantage of the fact that, for the first time, we have information available on both the single-particle optical properties of various microorganisms and a numerical radiative transfer model that is computationally efficient enough to permit extensive analysis of the effect of particle types and concentrations on underwater light fields. In addition, information on the optical properties of particles is used in the NASA-sponsored project on the analysis of photosynthetic rate and bio-optical components from ocean color imagery. The important aspect of the NASA project which is directly linked to this study of particle optics is that processes of photosynthesis and chlorophyll fluorescence in phytoplankton are modeled as a consequence of the fate of photons that are absorbed by the cells (Kiefer, 1994).

### *FINAL STATISTICAL INFORMATION*

#### *Key to references and conference presentations:*

*P* - Refereed publication

*PS* - Paper submitted

*PI* - Paper in preparation

*R* - Technical report

*C* - Presentation at technical conference

*IC* - Invited presentation at conference

*B* - Book or Chapter

#### A. LIST OF PUBLICATIONS

*P* - Stramski D., G. Rosenberg, and L. Legendre, 1993: Photosynthetic and optical properties of the marine chlorophyte *Dunaliella tertiolecta* grown under fluctuating light caused by surface wave focusing. *Mar. Biol.*, 115, 363-372.

*P* - Stramski D., and R. A. Reynolds. 1993. Diel variations in the optical properties of a marine diatom. *Limnol. Oceanogr.*, 38, 1347-1364.

*P* - Stramski, D., and M. Sedlak, 1994: Application of dynamic light scattering to the study of small marine particles. *Appl. Opt.*, 33, 4825-4834.

*P* - Dera, J., S. Sagan, and D. Stramski, 1994: Focusing of sunlight by sea surface waves: new results from the Black Sea. *Oceanologia*, 34, 13-25.

*B* - Kiefer, D. A., 1993: Modelling growth and light absorption in the marine diatom *Skeletonema costatum*. In: Towards a model of ocean biogeochemical processes, G. T. Evans and M. J. R. Fasham (editors), NATO ASI Series, Vol. I 10, Springer-Verlag, pp. 93-121.



B - Cullen, J. J., R. J. Geider, J. Ishizaka, D. A. Kiefer, J. Marra, E. Sakshaug and J. A. Raven, 1993: Towards a general description of phytoplankton growth for biogeochemical models. In: Towards a model of ocean biogeochemical processes, G. T. Evans and M. J. R. Fasham (editors), NATO ASI Series, Vol. I 10, Springer-Verlag, pp. 153-176.

B - Kiefer, D. A., 1994: Light absorption, fluorescence, and photosynthesis: *Skeletonema costatum* and field measurements. In: Ocean Optics, R. W. Spinrad, K. L. Carder, and M. J. Perry (editors), Oxford University Press, New York, pp. 165-188.

PS - Stramski, D., A. Shalapyonok, and R. A. Reynolds, 1994: Optical characterization of the oceanic unicellular cyanobacterium *Synechococcus* grown under a day-night cycle in natural irradiance. Submitted to Journal of Geophysical Research.

PS - Ondercin, D., C. A. Atkinson, and D. A. Kiefer, 1994: The distribution of bioluminescence and chlorophyll during the late summer in the North Atlantic: Maps and a predictive model. Submitted to Journal of Geophysical Research.

R - Stramski, D., 1994: Gas microbubbles: An assessment of their significance to light scattering in quiescent seas. In: Ocean Optics XII, SPIE Proceedings, in press.

R - Kiefer, D. A., C. A. Atkinson, and D. Ondercin, 1994: A bio-optical mapping procedure for the north Atlantic Ocean. In: Ocean Optics XII, SPIE Proceedings, in press.

R - Mobley, C. D., and D. Stramski, 1994: Influences of microbial particles on oceanic optics. In: Ocean Optics XII, SPIE Proceedings, in press.

#### B. NUMBER OF GRADUATE STUDENTS

None under this contract

#### C. PATENTS

None

#### D. PRESENTATIONS/BRIEFINGS

C - Kiefer, D. A.: Mapping the distribution of bioluminescence and chlorophyll from ocean color imagery. Symposium on Marine Light - Mixed Layer Project, University of Rhode Island, 1993.

IC - Kiefer, D. A.: Mapping the distribution of chlorophyll in the North Atlantic from ocean color imagery. Invited talks at the Scripps Institution of Oceanography and Lamont-Doherty Geological Observatory of Columbia University, 1993.

IC - Kiefer, D. A.: Modelling the distribution of phytoplankton in the Southern Ocean. Invited talk at the Bigelow laboratory for Ocean Sciences, 1993.

*IC* - Kiefer, D. A.: Alternative approaches to the productivity algorithm. Invited talk at NASA's SeaWiFS Workshop, Brookhaven, New York, January 1994.

*C* - Kiefer, D. A.: Mapping bio-optical properties of the upper ocean from space. Contributed seminar at the Office of Naval Research, Arlington, Virginia, January 1994.

*C* - Berwald J., D. Stramski, C. D. Mobley, and D. A. Kiefer: A study of the relationship between the mean cosine for irradiance and inherent optical properties of the ocean. Contributed presentation at 1994 Ocean Sciences Meeting, AGU and ASLO, San Diego, California, February 1994.

*IC* - Kiefer, D. A. and R. A. Reynolds: Laboratory and field studies of the relationship between natural fluorescence and photosynthetic rate. Invited presentation at 1994 Ocean Sciences Meeting, AGU and ASLO, San Diego, California, February 1994.

*C* - Stramski D.: Gas microbubbles: An assessment of their significance to light scattering in quiescent seas. Contributed presentation at SPIE International Conference, Ocean Optics XII, Bergen, Norway, June 1994.

*C* - Kiefer, D. A., C. A. Atkinson, and D. Ondercin: A bio-optical mapping procedure for the north Atlantic Ocean. Contributed presentation at SPIE International Conference, Ocean Optics XII, Bergen, Norway, June 1994.

*IC* - Kiefer, D. A.: A bio-optical model of phytoplankton and the 3-dimensional mapping of chlorophyll from satellite ocean color imagery. Five invited talks in Bremerhaven, Hamburg (Germany), Southampton, Plymouth (U.K.) and Ispra (Italy), July-September 1994.

#### E. SERVICE ON COMMITTEES/PANELS

Stramski served on the Ph.D. dissertation committee of two graduate students at the University of Southern California, and Kiefer served on such a committee of twelve graduate students. Kiefer has also been a Head of Marine Biology Research Section at the Department of Biological Sciences at USC.

#### F. HONORS/AWARDS

The presentation entitled "A study of the relationship between the mean cosine for irradiance and inherent optical properties of the ocean" by J. Berwald, D. Stramski, C. D. Mobley, and D. A. Kiefer at 1994 Ocean Sciences Meeting, AGU and ASLO, San Diego, (February 1994), was named Outstanding Student Paper.